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Filamin C Mutations and Susceptibility to Chemotherapy-Induced Dilated Cardiomyopathy: Exploring the Role of Genetic Predisposition in Adriamycin Toxicity

Abstract

Dilated cardiomyopathy (DCM) is defined as ventricular dilation and systolic dysfunction in the absence of ischemic disease or abnormal loading conditions. Mutations in *FLNC*, which encodes the intracellular structural protein Filamin C (Fil C), are increasingly associated with inherited cardiomyopathy. Filamin C plays a key role in sarcomere anchoring through its actin-binding domain, and more than 300 variants of *FLNC* have been described, many of which are nonsense or frameshift mutations. Doxorubicin, a widely used chemotherapeutic agent, has an established link to chemotherapy-induced DCM. However, the impact of doxorubicin on Filamin C-mutated cardiomyocytes remains underexplored. This study investigated doxorubicin-induced cardiotoxicity in induced pluripotent stem cell–derived cardiomyocytes (iPSC-CMs) carrying a Filamin C mutation compared with normal iPSC-CMs.

Fil C mutant and normal iPSC-CMs were differentiated and cultured in 24-well regular and micro electrode array (MEA) plates, respectively, and were either treated with 300 nM doxorubicin or left untreated. Metrics such as time-to-peak, relaxation time, and peak-to-peak times were analyzed using MUSCLEMOTION (MM), while other metrics, including beat period, conduction velocity, beat amplitude, and field potential duration, were obtained from the MEA plate via MAESTRO. RNA was harvested from both plates for mRNA expression profiling, and genomic DNA was extracted to confirm the Filamin C mutation.

MM analysis showed that doxorubicin significantly reduced relaxation and peak-to-peak times in both normal and mutant iPSC-CMs. Treated normal IPS-CMs demonstrated a time-to-peak increase at 24 hours, followed by a decline below control levels by 48 hours, with the greatest difference observed at 96 hours. Fil C-mutated cells exhibited their largest time-to-peak difference at 72 hours, after which the values of the treated and untreated groups converged. MAESTRO data analysis revealed that in normal iPSC-CMs, beat amplitude increased through 48 hours before declining to undetectable levels at 96 hours, although MM continued to detect contraction until 120 hours. Conduction velocity and beat period progressively declined in treated groups, while field potential duration decreased after 48 hours. Fil C-mutated cells repeatedly failed to reach a viable state on MEA plates, preventing direct comparison with MAESTRO software.

These results demonstrate that doxorubicin negatively affected both normal and Fil C-mutated IPS-CMs. Doxorubicin appeared to induce early hypercontractility in the MEA plate, followed by eventual failure at 96 hours. However, MM was still able to detect activity in the MEA plate until termination at 120 hours. While MM analysis revealed measurable differences in timing between groups, limitations in Fil C cell viability restricted MEA plate MAESTRO assessment. Ongoing mRNA expression profiling will provide further insight into differential gene expression and pathways contributing to doxorubicin-induced cardiotoxicity in the setting of Fil

C-mutation. Future efforts will prioritize optimizing Filamin C MEA culture conditions to enable more direct comparisons. mRNA expression data analysis is in process.